Successful introduction of infant dried blood spot (DBS) PCR testing in Botswana's PMTCT program

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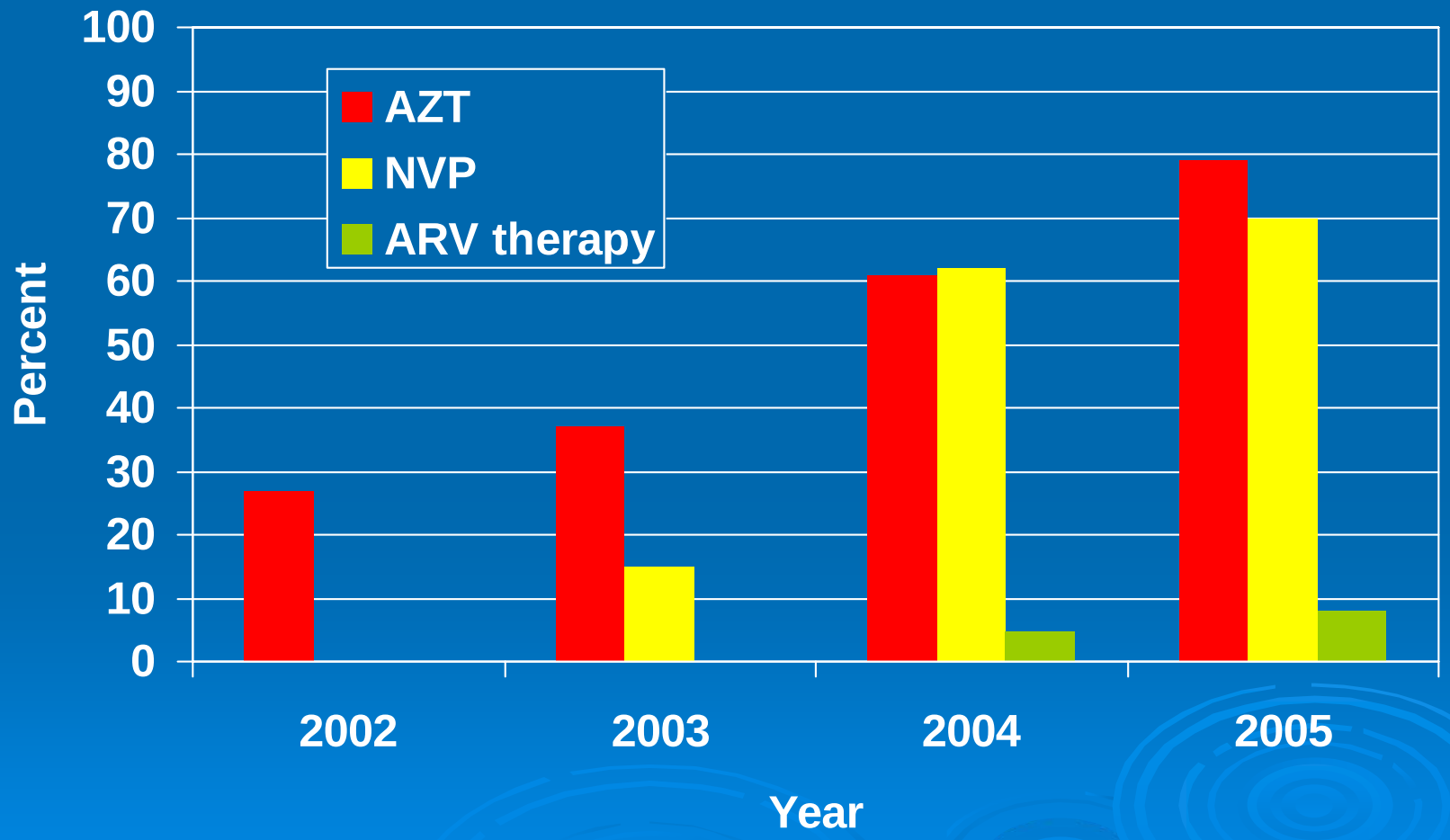
Botswana – prevention of mother to child transmission (PMTCT)

- 37.1% of pregnant women HIV+
- PMTCT started 1999, national since 2001
- Interventions
  - Routine HIV testing during pregnancy
  - ARV therapy if CD4<200
  - AZT 12 wks to mother, 4 wks to baby
  - SD NVP mother & baby
  - Infant formula x 12 months
- New child health card has PMTCT interventions, infant feeding, & HIV testing

Percent of all HIV-positive women receiving PMTCT interventions, Botswana national PMTCT program, 2002-2005



Botswana – ARV therapy

- ARV program started 2001
- National expansion complete 2004
- ~60,000 people on therapy (3,000 are children)
- Pediatric ARV available at large sites, expanding
- 2005 pediatric ARV guidelines
  - <12 months – ARV regardless of CD4
  - >12 months – ARV based on CD4, clinical criteria

Infant HIV transmission & followup within PMTCT program

- Based on detailed PMTCT uptake data, we estimated about 6% of HIV-exposed infants born in Francistown in 2005 were HIV infected
- Transmission within national program not documented previously
- Infant PCR on whole blood available since 2001 but only at a few sites, problems in lab with turnaround time, sample storage

Pilot of PCR on DBS

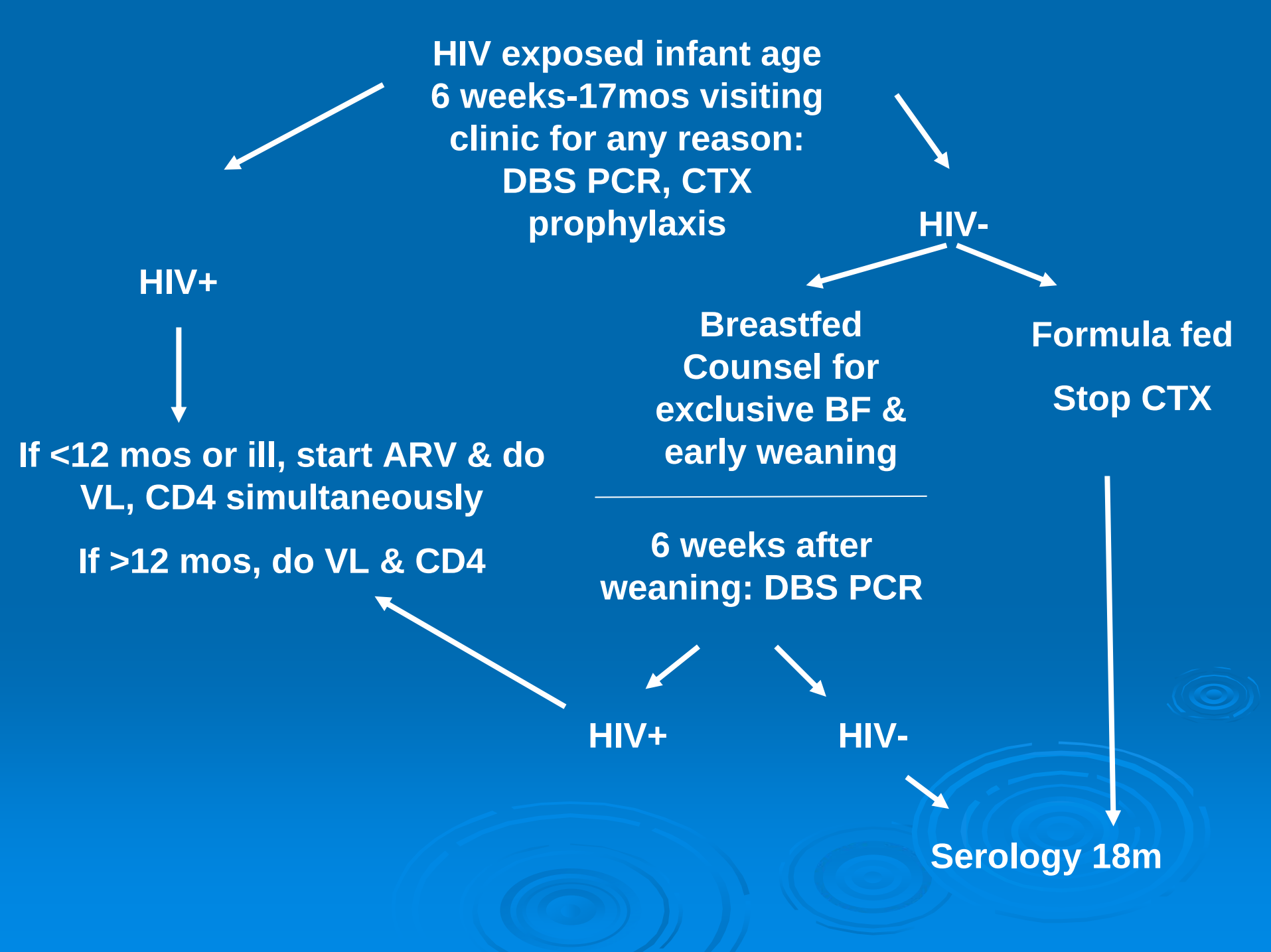
- Dried blood spots expected to solve storage, transport, and blood collection problems and allow wide access to early diagnosis
- Pilot objectives:
  - Determine feasibility of DBS collection at government clinics during routine infant care
  - Identify and solve problems with sample collection
  - Establish lab QA system
  - Document rates of HIV infection among infants
  - Track infants diagnosed early to determine if early diagnosis leads to early treatment

Pilot of PCR on DBS – clinical sites

- Pilot lasted June – December 2005
  - Francistown: 10 clinics & regional referral hospital
  - Gaborone: Botswana-Baylor Children's Center of Excellence
- 250 nurses, midwives, doctors trained in DBS collection
  - 1 day classroom training (Review of HIV testing principles, ARV guidelines, pre & post-test counseling, paperwork, new child health card, DBS)
  - Several days hands-on training at each site
  - 4-5 infants per provider required for proficiency at DBS collection

Pilot of PCR on DBS - lab

- Dedicated technician at national HIV reference lab doing all testing (can do 48 samples/day)
- Roche Amplicor 1.5 with manual extraction
- Samples transported by DHL
- Results transmitted by fax


HIV exposed infant age 6 weeks-17mos visiting clinic for any reason: DBS PCR, CTX prophylaxis
HIV+
HIV-
If <12 mos or ill, start ARV & do VL, CD4 simultaneously
If >12 mos, do VL & CD4
Breastfed Counsel for exclusive BF & early weaning
Formula fed
Stop CTX
6 weeks after weaning: DBS PCR
HIV+
HIV-
Serology 18m

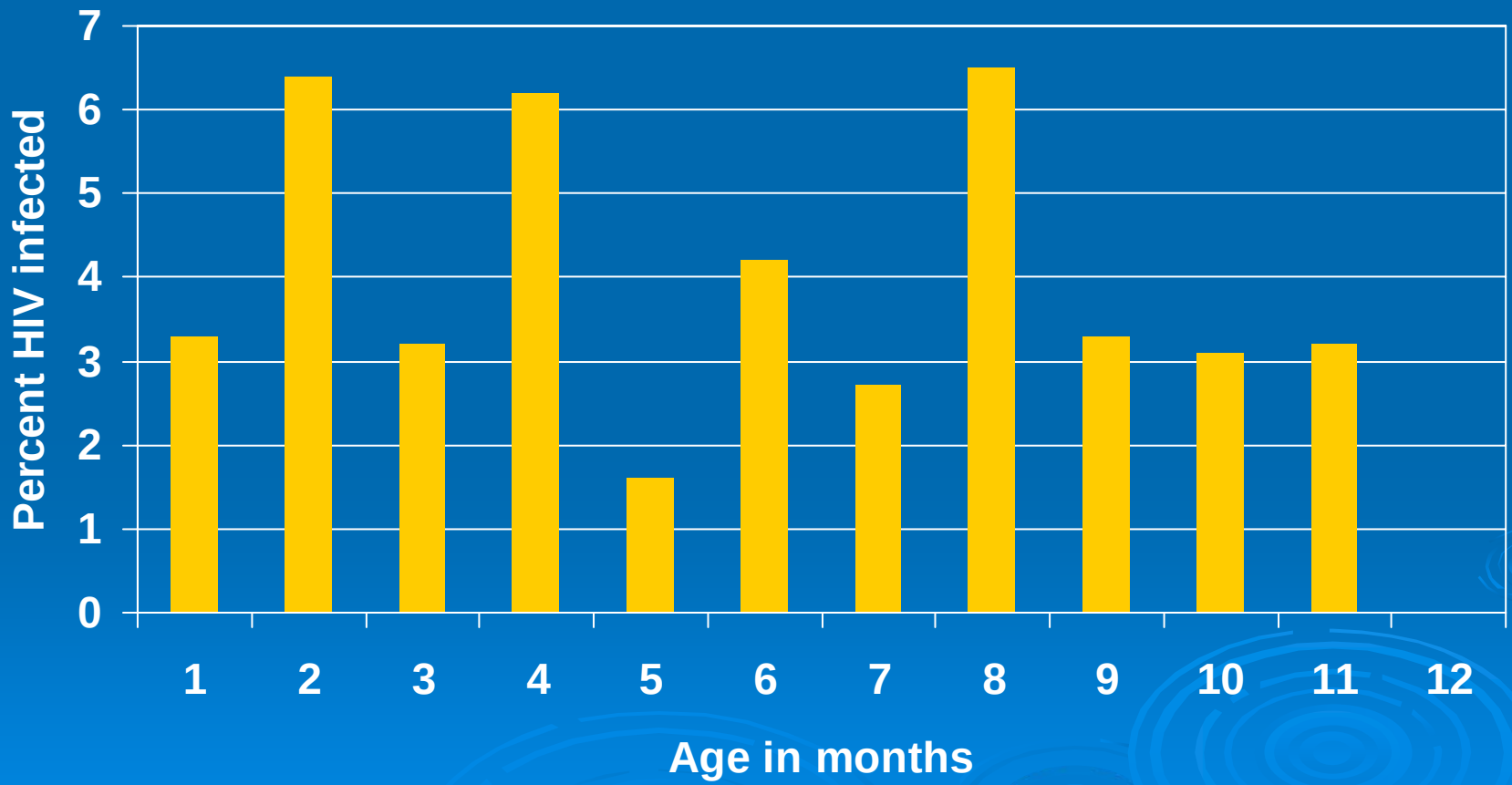
Results - pilot of PCR on DBS

- 1917 HIV-exposed infants tested, age 6 weeks-18 months

- Overall 6.7% HIV+
  - In clinics 4.4% HIV+ (all outpatients, N=1356)
  - At hospital 12% HIV+ (inpatients and outpatients, N=561)

- Among 1356 outpatient infants tested in clinics
  - 99% received at least 1 intervention for PMTCT
  - 98% formula-fed
  - 81% of babies age 3-12 months on CTX
  - 81% of caregivers received results of infant test

Percent HIV-infected by age in months, outpatient infants tested by DBS PCR -- Botswana, 2005 (n=1412)



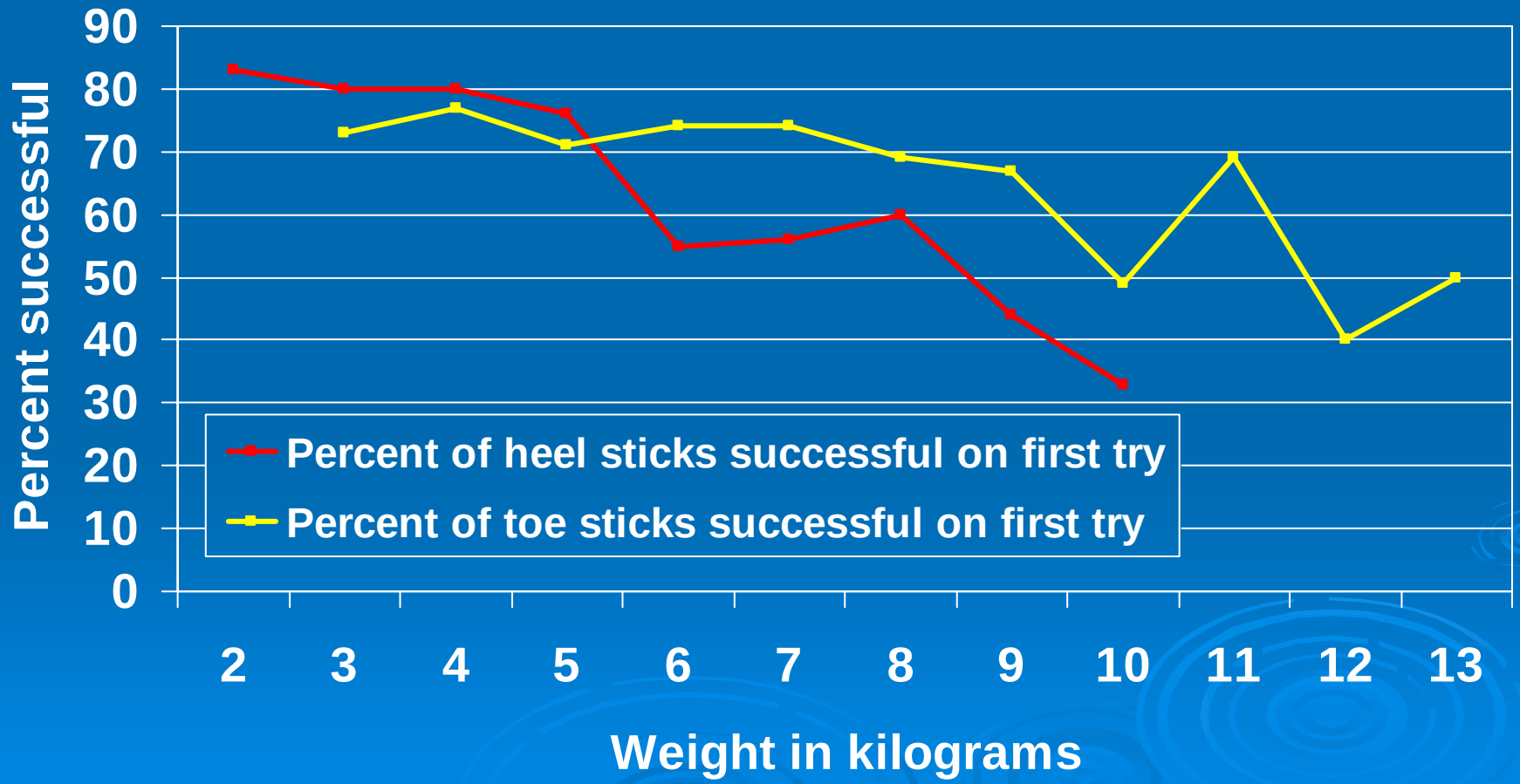
Percent of infants HIV-infected by PMTCT interventions received, DBS PCR pilot – Botswana, 2005

Regimen	N	% expected positive	% actually positive
Nothing	13	35-40	31
Nothing to mother, AZT/NVP/formula for baby	22	12-20	9
AZT to mother (median 49 days, many also rec'd NVP)	1108	2-8	3.7
ARV therapy to mother	170	<1	0.7

Results - pilot of PCR on DBS

- Operationally,
  - DBS worked great
  - DBS acceptable to staff and mothers: >90% of the HIV-exposed infants <12 months in Francistown were tested during the 6 month pilot
  - Nurses and midwives were equally successful at collecting DBS, 67% of samples collected by nurses
  - 73% of infants were stuck only once
  - Heel sticks worked best for age 1-4 months/<6kg
  - Toe sticks worked best for age 4-10 months/<10kg
  - Finger sticks may be needed for older babies (>10kg)

Percent of infant sticks successful on the first try, by site of blood collection and weight -- dried blood spot HIV PCR pilot study – Botswana, 2005 (n=1314)



Results – pilot of PCR on DBS

- No problems in the lab
- All positive results confirmed by second test on same sample
- No false positives detected in ARV clinic
- CDC QA samples 100% correctly tested
- 1.7% of samples rejected by lab
  - Labeling errors most common reason
  - Sample quality rarely a problem
- Average turnaround time = 9 days from collection to receiving result in clinic

HIV-infected infants

- 38 HIV-infected infants identified in FT clinics during pilot period
  - 34 caregivers (90%) received results
- 22 (58%) were seen in ARV clinic by end of January 2006
  - 17 started therapy
  - 3 evaluation not complete
  - 2 did not need therapy yet
- 3 died before therapy
- 3 moved out of the area
- 10 no followup

Conclusions

- DBS collection easily integrated into routine infant care in clinics
- One-on-one training required for proficiency
- Minor problems to address before expansion
- PMTCT program functioning well, very low rates of infection among infants who received PMTCT interventions
- Infants tested often received early therapy, but there was a high rate of loss to followup

Sample collection issues to address before national rollout

- Biggest problem was labeling
  - Use names, not just numbers
  - Cards attached to forms like in US newborn screening?
  - Stickers? Bar codes?
- A few other minor issues
  - Blood from femoral stick/syringe most likely to produce poor sample due to clotting or hemolysis, should be discouraged unless necessary for other tests

Next steps

- Training video w/Roche (*not* Botswana specific)
- Supply procurement!
- Discussions with MOH:
  - Labeling issues
  - Supply storage, distribution
- National rollout using dedicated training teams will start later this year, funded by USG
- Discussions with industry:
  - DBS cards attached to forms?
  - Purchase supplies needed to test x babies in one box?

Acknowledgements

- CDC infant diagnosis working group
- CDC-Botswana staff
- Staff of Francistown City Council, Nyangabgwe Hospital, Botswana-Baylor Children's Center of Excellence
- Botswana-Harvard Partnership laboratory
- Botswana National PMTCT Program
- All the mothers who eagerly brought their babies for testing